

Fine needle aspiration cytology in the diagnosis of bone tumours

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Abstract

Fine needle aspiration (FNA) cytology was performed on 62 primary and 22 metastatic bone tumours. Histological correlation was available in all 62 primary bone tumours, which included 7 benign and 53 malignant tumours. The most common of the malignant tumours were Ewing's sarcoma, followed by giant cell tumour, osteosarcoma and chondrosarcoma. The benign tumours included 4 enchondromas, 1 chondromyxoid fibroma and 2 osteochondromas.

The overall cytodiagnostic accuracy in primary malignant bone tumours was 86.9%, the specificity was 100% and cytological categorization of tumours was possible in the majority of cases. This eliminated the need for bioptic confirmation. Our results indicate that FNA cytology is a valuable diagnostic tool in bone tumours.

Key words: Fine needle aspiration cytology, bone tumours.

INTRODUCTION

The fine needle aspiration (FNA) cytology technique has been applied successfully to the distinction of primary from metastatic bone tumours and the categorisation of primary bone tumours. Occasionally it also provides evidence against a diagnosis of a bone tumour.¹⁻⁵ Although bone is a hard tissue and presents a natural barrier to FNA cytology, most malignant bone tumours cause bone destruction either in the form of lytic areas or pathological fractures, or erode through the bone and produce soft tissue swellings that can be needed. It is therefore possible to needle the majority of bone tumours and make a preoperative cytodiagnosis. In this paper we present a cytological analysis of 84 bone tumours that were subjected to fine needle aspiration.

MATERIALS AND METHODS

The material for the study was obtained from 84 patients who were selected from several hundred patients with primary or metastatic bone tumours presenting at the Lok Nayak Jai Prakash Narain Hospital between the years 1985 and 1991 and referred to the cytopathologist for FNA cytology. Detailed clinical and radiological assessment of the lesion was done in each case. In palpable lesions aspiration was performed with a needle of external diameter 0.6 to 0.7 mm (23 or 24 gauge). Non-palpable lesions were aspirated with the help of radiographs or fluoroscopic guidance. In cases where a thinned out cortex

had to be penetrated, 0.9 mm needles were used. The needle was attached to a 20 c.c. syringe mounted on a handle for single hand grip. Six to eight smears of the aspirate were prepared in each case on clean glass slides, two of which were wet fixed in ethanol for Papanicolaou or special staining techniques. The rest were air dried, fixed in methanol and stained with May Grunwald Giemsa (MGG) staining technique. Cytological features were incorporated with the clinical and radiological features to arrive at the final cyto-diagnosis. In all 62 primary bone tumours, open surgical biopsy, excision of tumour or amputation of the limb provided material which was processed routinely for histopathological study.

Considering all malignant bone lesions as true positives, the cyto-diagnostic sensitivity (ability to detect true positives), specificity (ability to correctly identify true negatives) and the overall cyto-diagnostic accuracy rate (percentage of cyto-diagnosis corresponding to histodiagnosis) were determined.

RESULTS

A correlation of the cytodiagnosis with the histodiagnosis in the 62 primary bone tumours is shown in Table 1. The overall accuracy of cytodiagnosis was 86.9% and the specificity 100%. The cytodiagnostic accuracy for benign tumours was 42.9% and for malignant tumours 92.6%. The sensitivity of FNA cytology for the diagnosis of primary malignant tumours was 98.1%.

TABLE 1: Cytodiagnosis correlated with histodiagnosis in primary bone tumours

Histodiagnosis	Number	Cytodiagnosis	Number
Enchondroma	4	Enchondroma Inadequate	2 2
Chondromyxoid fibroma	1	Chondroid tumour	1
Osteochondroma	2	Osteochondroma Uncharacteristic cytology	1 1
Ewing's sarcoma	15	Ewing's sarcoma	15
Osteosarcoma	9	Osteosarcoma	9
Giant cell tumour	13	Giant cell tumour Malignant fibrous histiocytoma	12 1
Chondrosarcoma	6	Chondrosarcoma Chondroid tumour	5 1
Synovial sarcoma	2	Synovial sarcoma ? Fibrosarcoma/ ? Monophasic synovial sarcoma	1 1
Myeloma	2	Myeloma	2
Malignant fibrous histiocytoma	3	Malignant fibrous histiocytoma Poorly differentiated sarcoma	2 1
Poorly differentiated sarcoma	3	Poorly differentiated sarcoma	3
Chordoma	1	Chordoma	1
Total	61	Total	61

Cytological features

Adequately cellular smears from 2 *enchondromas* showed cartilaginous fragments with small rounded chondrocytes, some within lacunae. The cells were often not clearly discernable owing to thickness and strong staining of cartilage matrix which was abundant and gave a background metachromatic staining reaction with MGG. This characteristic appearance of the chondroid stroma was lost with the Papanicolaou staining technique though the lacunae were often better discerned.

The case of *chondromyxoid fibroma* showed a few chondroid cells with smudgy nuclei, occasional binucleated forms and a few fibroblastic cells. The cellularity was suboptimal and a cytological diagnosis of a benign chondroid tumour was made.

In one of the two cases of *osteochondroma*, smears showed inadequate cellularity and no

cytological opinion was possible. The other case showed scattered spindled out stromal cells, chondrocytes, osteoblasts and occasional osteoclastic giant cells. The osteoblasts were dispersed or in short rows, groups or clusters, and were medium-sized, round or polygonal with moderate amount of dense blue cytoplasm and eccentric vesicular round nuclei. A few cells showed small cytoplasmic vacuoles. Osteoclastic giant cells showed five to ten nuclei and cells of probable fibroblastic type with fibrillary cytoplasmic processes appeared to be maturing into chondrocytes and osteoblasts. Correlating cytological with radiological features, a diagnosis of osteochondroma was given.

Smears in *chondrosarcoma* were poorly or moderately cellular and showed abundant deeply staining metachromatic myxoid or hyaline matrix enclosing single cells or small cell groups, occasionally in lacunae (Fig. 1). Binucleated

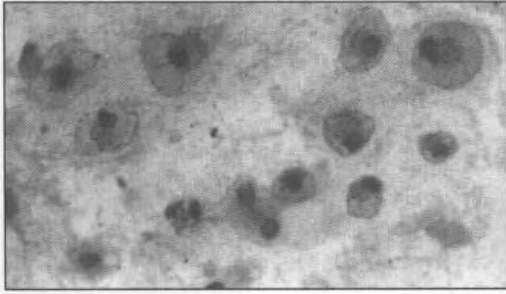


FIG. 1: Smear from chondrosarcoma showing single cells, some in lacunae, in a myxoid background. MGG $\times 300$.

and trinucleated cells were present and there was mild nuclear pleomorphism. Two cases showed moderate pleomorphism. A few osteoclastic giant cells were scattered. One case that was cytologically diagnosed as a mesenchymal chondrosarcoma showed dissociated small round cells measuring three to four times the size of red blood cells. A perivascular survival pattern was prominent, matrix was sparse and occasional chondroid cells with abundant cytoplasm stood out in stark contrast to the round cells (Fig. 2).

Smears from *osteosarcoma* showed high cellularity with dissociated single osteoblastic cells and occasional short rows or clusters of cells (Fig. 3). Nuclear pleomorphism and cells with bizarre mitosis were seen in some cases. Multinucleated tumour cells containing three to five nuclei (Fig. 3) or giant cells with bizarre lobulated nuclei were seen in three cases. Osteoclast-like giant cells were common and the background of the smears showed a granular appearance (Figs. 3 & 4) due to the spread out

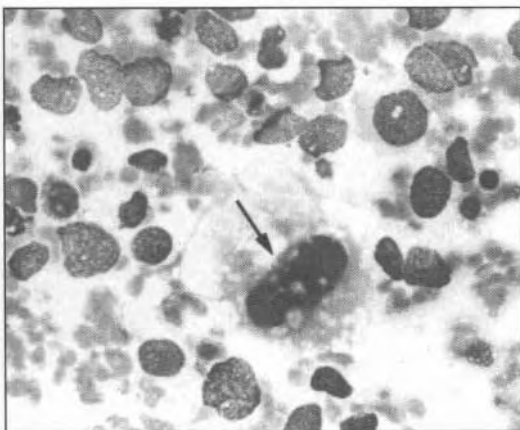


FIG. 2: Scattered round cells and one cell with abundant cytoplasm (arrow) in mesenchymal chondrosarcoma. MGG $\times 400$

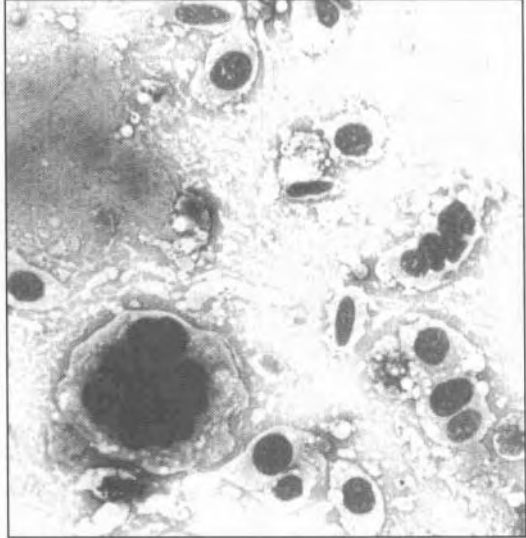


FIG. 3: Dissociated and short rows of osteoblasts with one bizarre multinucleated cell in osteosarcoma. MGG $\times 400$

matrix that stained pink with MGG. The MGG stain was advantageous in bringing out the characteristic appearance of the matrix which was not appreciated with the Papanicolaou staining technique. Fragments of osteoid were also occasionally discernible which incorporated osteoblastic cells (Fig. 5). One case showed a predominantly chondroid differentiation.

Smears from *Ewing's sarcoma* were highly cellular consisting of dissociated, rosette-like or loose clusters of small round cells (Figs. 6 & 7) about twice the size of red blood cells. Cells with intact cytoplasm showed delicate cytoplasmic vacuolation (Fig. 7) that was

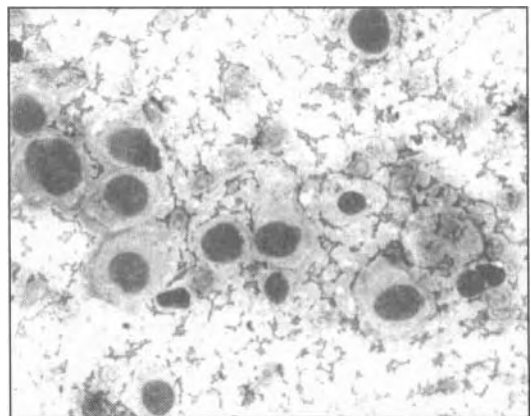


FIG. 4: Scattered well-differentiated osteoblasts in a background of granular material in osteosarcoma. MGG $\times 400$



FIG. 5: Dark staining fragment of osteoid incorporating osteoblasts in osteosarcoma. MGG x 250

demonstrated to be glycogen by special staining techniques. Perivascular survival pattern was prominent and pleomorphism and mitotic activity insignificant.

Cellularity was moderate in *giant cell tumours* and consisted of ovoid to plump stromal cells that were dissociated or clustered (Fig. 8). Many cells with cytoplasmic vacuolation and a few cells with elongated fibrillary cytoplasm were present. Occasional binucleated cells and cells in mitosis were seen. Multinucleated giant cells (Fig. 8), some with more than 20 nuclei were present and a perivascular survival pattern of stromal cells was present in some of the cases.

Smears from *chordoma* showed a swirling pattern of abundant metachromatic fibrillary mucoid matrix with enmeshed tumour cells (Fig. 9). The tumour cells were large and physaliferous with abundant vacuolated to clear cytoplasm and vesicular nuclei with a few binucleated forms. Also seen were small round cells and a few plump spindle cells. Of two *synovial sarcomas*, one was biphasic and showed dissociated oval to

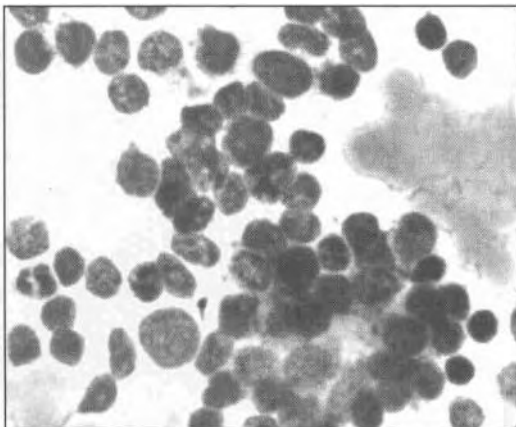


FIG. 6: Rosette-like clusters and dissociated small round cells in Ewing's sarcoma. MGG x 600

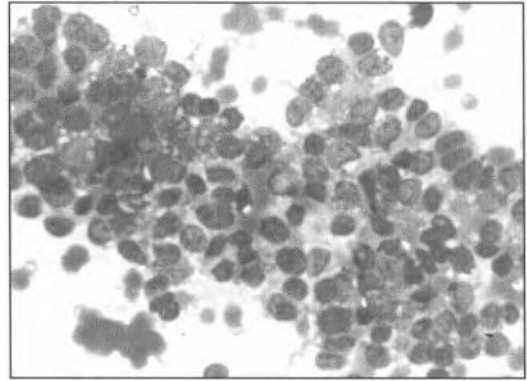


FIG. 7: Clusters of small round cells in Ewing's sarcoma, some with delicate cytoplasmic vacuolation. MGG x 400

spindle shaped cells with pale delicate cytoplasm and a few clustered cells with an epithelioid appearance. A perivascular pattern was present. Monophasic synovial sarcoma showed predominantly spindle cells that were dissociated or in fascicles and in this case distinction from a low grade fibrosarcoma was not possible.

Smears from *malignant fibrous histiocytoma* showed moderate cellularity with an admixture of spindle cells, histiocytic cells with foamy cytoplasm and pleomorphic multinucleate giant cells, some of *Touton* type. The spindle cells were dissociated and clustered and occasional foci of *storiform* pattern could be discerned. Some of the histiocytic cells showed ingested acellular material as well as intact cells. The giant cells and the histiocytes were scattered and mitotic activity and pleomorphism were present. Fat *and/or* hemosiderin could be demonstrated with special staining techniques in some of the histiocytes and multinucleated giant cells.

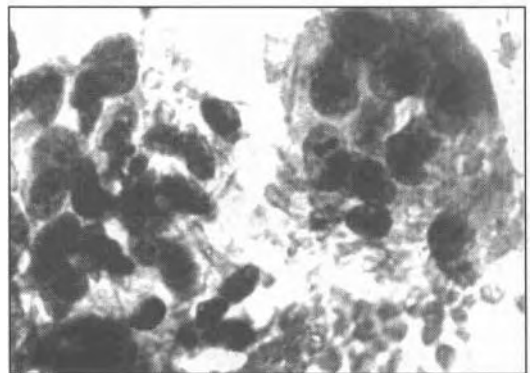


FIG. 8: Stromal cells and multinucleated giant cell in giant cell tumour. MGG x 400

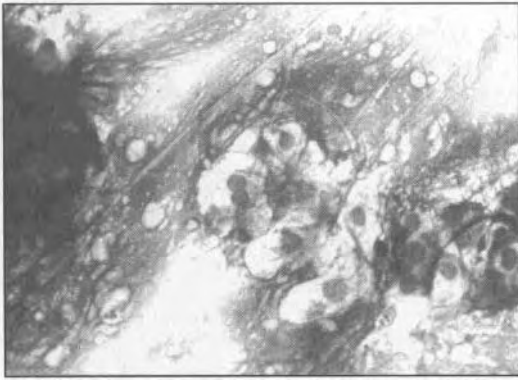


FIG. 9: Metachromatic fibrillary matrix surrounding physaliferous tumour cells in chordoma. MGG \times 300

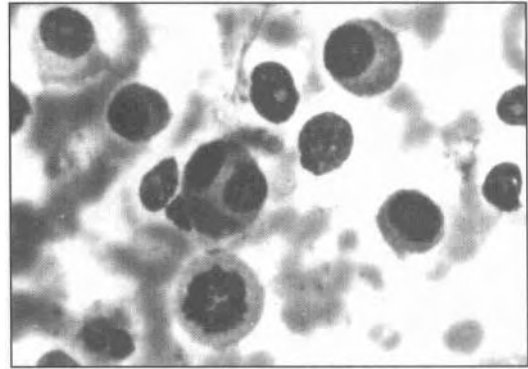


FIG. 10: Smear from myeloma showing plasma cells and myeloma cells with one cell in mitosis. MGG \times 400

Smears from both cases of *myeloma* showed numerous scattered and clustered plasma cells and myeloma cells (Fig. 10).

Three cases of *poorly differentiated sarcoma* showed dissociated spindle cells, plump cells and tumour giant cells with prominent pleomorphism and mitotic activity.

Cytological diagnosis in 22 *metastatic bone tumours*, is given in Table 2. These included 15 carcinomas, 3 neuroblastomas and 4 sarcomas. Three of the adenocarcinomas showed intestinal type of columnar cells and mucous secreting cells including goblet cells that suggested a primary in the gastrointestinal tract. This however could not be confirmed as all three patients were lost to follow up. In two metastatic prostatic adenocarcinomas and breast carcinomas as well as one bronchogenic small cell undifferentiated carcinoma, the primary site of malignancy was known and the cytological picture was compatible with metastases from these sites. In three cases the primary was occult and the cytology could

only provide a broad diagnosis of metastatic adenocarcinoma.

All four *metastatic renal cell carcinomas* showed a characteristic clear cell population (Fig. 11) and lipid could be demonstrated on special stains. In two of these the primary was occult at the time of bony metastasis.

The orbit was the site of metastases in all three cases of *neuroblastoma*. Cytological smears showed round cells with hyperchromatic nuclei, mild *anisocytosis* and anisonucleosis. Cells were also aggregated in rosette-like pattern with fibrillary material in the center (Fig. 12). Detailed clinical and investigative *workup* of these patients confirmed the cytological diagnosis of neuroblastoma.

The two cases of *metastatic fibrosarcoma* showed dissociated and clustered spindle cells some of which were associated with acellular *stroma-like* material (Fig. 13) that exhibited staining characteristics of collagen. The metastatic neurofibrosarcoma showed a pleomorphic picture with many bizarre

TABLE 2: Cytological diagnosis in 22 metastatic bone tumours

Cytological diagnosis	No. of cases
Metastatic gastrointestinal adenocarcinoma	3
Metastatic breast carcinoma	2
Metastatic prostatic adenocarcinoma	2
Metastatic adenocarcinoma (site not specified)	3
Metastatic renal cell carcinoma	4
Metastatic bronchogenic carcinoma	1
Metastatic neuroblastoma	3
Metastatic fibrosarcoma	2
Metastatic neurofibrosarcoma	1
Metastatic embryonal rhabdomyosarcoma	1
Total	22

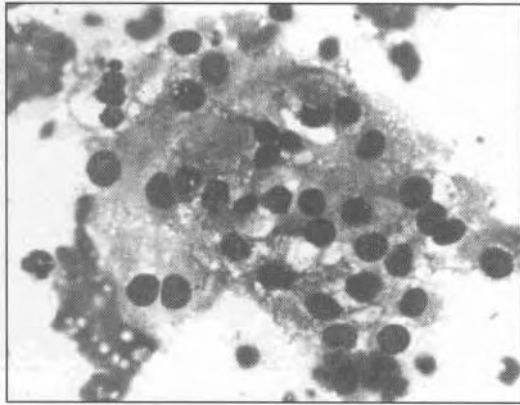


FIG. 11: Cluster of clear cells in metastatic renal cell carcinoma. MGG \times 300

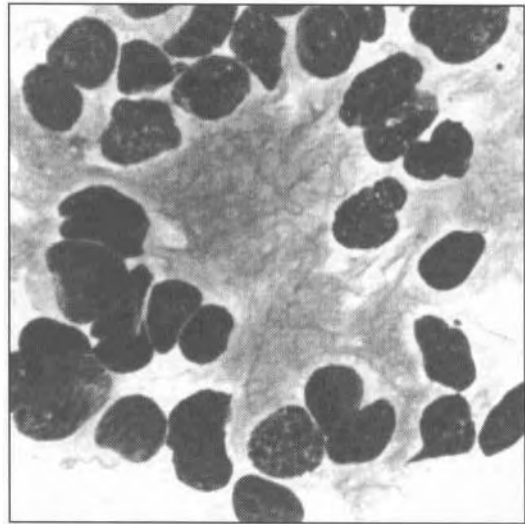


FIG. 12: Tumour cells in rosette-like pattern with central fibrillary material in metastatic neuroblastoma. MGG \times 600

multinucleated giant cells, abnormal mitosis, plump cells, spindle cells and only occasional clusters of cells with elongated nuclei and tapering fibrillary cytoplasm suggestive of neural origin. All three patients had undergone prior limb amputation for the primary tumour.

The metastasis of *embryonal rhabdomyosarcoma* occurred in the orbit. Smears showed round cells with mild to moderate cellular pleomorphism and a prominent perivascular pattern. Focal arrangement of nuclei in wreath pattern and scattered myoblastic cells with abundant deeply staining cytoplasm enabled a cytodiagnosis of embryonal rhabdomyosarcoma. The primary which had been occult was later discovered in the nasopharynx and the cervical lymph node metastasis was subjected to excision and histopathological confirmation. Sections showed a pseudoalveolar pattern of tumour cells some of which showed myoblastic differentiation while others showed a wreath-like arrangement of nuclei.

Histological confirmation was not done on the remaining 21 metastatic bone tumours. All but one of the metastatic bone lesions were osteolytic or had caused pathological fractures. In one case the metastatic prostatic tumour presented as a markedly osteosclerotic lesion in the left humerus. FNA could however be done from the soft tissue that was involved by the tumour. Of the primary malignant bone tumours, all but six osteosarcomas presented with either osteolytic lesions or pathological fractures. Some of the osteolytic lesions had intact or thinned out cortex which could be penetrated, while others had destroyed the cortex.

DISCUSSION

The diagnosis of bone tumours is always made

by integrating the morphological features with the clinico-radiological picture. Our results indicate that FNA cytologic study guided by clinical and radiological features can be a valuable tool in the diagnosis and also categorization of primary and metastatic bone tumours. This has also been the experience of many other workers.¹⁻⁷

On the whole, benign tumours showed suboptimal cellularity rendering cytological diagnosis more difficult than in malignant tumours where the smears were richly cellular. Ewing's sarcoma was the most common primary bone tumour encountered. Differentiation of Ewing's sarcoma from other round cell tumours such as embryonal rhabdomyosarcoma, neuroblastoma and lymphoma may be difficult in some cases and immunocytochemical markers and electron microscopy may be necessary.⁸⁻¹²

Smears from giant cell tumour, the next most common tumour, showed mononuclear cells and multinucleated giant cells.¹³ The cytology of giant cell tumours has been well described.^{1,14} On a purely cytological basis it may be difficult to distinguish giant cell tumour from other giant cell lesions of bone such as aneurysmal bone cyst and giant cell reparative granuloma.

The osteoid-like material seen in MGG stained smears was helpful in the cytological distinction of osteogenic tumours. Although the cellular pleomorphism was variable, by incorporating the clinical, radiological and cytological features, a 100% accurate diagnosis could be obtained which enabled planning and execution of radical surgery without any delay. This has also been

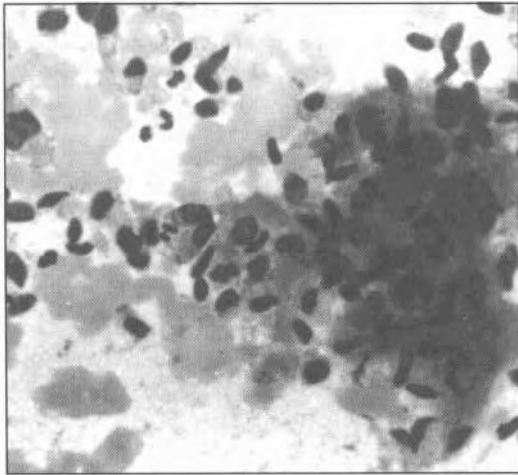


FIG. 13: Smear from metastatic fibrosarcoma showing dissociated and clustered spindle and ovoid cells. Cluster on the right is associated with stroma-like material. MGG \times 300

the experience of Walaas and Kindblom and

The morphology in chondroid tumours was also sufficiently distinctive with abundant extracellular metachromatic cartilaginous matrix in MGG stained smears. The cellularity however was scanty in some of these tumours and it may sometimes be difficult to distinguish benign chondroid tumours from low grade chondrosarcoma and high grade osteosarcoma from chondroblastic osteosarcoma.⁶

Koivuniemi and Nickels,¹⁸ Agarwal and Wahal¹⁹ and Kindblom²⁰ described the cytological picture in synovial sarcoma. The biphasic type may contain well developed gland-like structures²⁰ and clusters of epithelioid cells²¹ in addition to spindle cells. The monophasic variety may not be cytologically distinguishable from other spindle cell tumours like in one of our cases.

The cytological features of malignant fibrous histiocytoma have been well described and documented.²² The two main cell types i.e. the mononucleated or multinucleated, large, polymorphic, often bizarre histiocyte-like cells with phagocytic material and the fibroblast-like cells²² were seen in our aspirates from both cases of malignant fibrous histiocytoma enabling correct identification.

The cases of myeloma and the solitary case of chordoma also showed characteristic cytomorphology. All the three cell types, namely the large physaliferous cells with bubbly cytoplasm, small round cells and short spindle

cells^{23,24} which compose chordoma were seen in this case, enabling easy distinction from chondroid tumours, clear cell carcinoma and mucus secreting adenocarcinoma.

In metastatic bone tumours also our results with FNA cytology were satisfactory. In the majority of these lesions, the cytological features indicated the broad type of malignancy e.g. adenocarcinoma, small cell carcinoma, sarcoma etc. which when coupled with the knowledge of the primary tumour and review of the sections of the excised primary tumour, rendered diagnostic bone biopsy unnecessary. In some of the lesions such as metastatic clear cell carcinoma of kidney, metastatic embryonal rhabdomyosarcoma, metastatic neuroblastoma and metastatic gastrointestinal adenocarcinoma the primary lesions were occult and the cytological picture was sufficiently characteristic to enable identification of the primary tumour.

The preoperative investigation and evaluation of bone tumours present major challenges for clinicians, radiologists and pathologists. Diagnostic biopsy prior to radical surgery may entail technical problems and has not always been preferred by clinicians, some of whom have argued that the surgical removal of a bone tumour can be done without a prior biopsy, provided the clinical and radiologic findings are highly suggestive.^{6,15} Others however, consider a preoperative morphologic diagnosis to be essential before the planning and execution of radical surgery. From our study we conclude that FNA cytology is a simple, quick and excellent diagnostic adjunct in primary and metastatic bone tumours, giving high reliability and accuracy when used in a multidisciplinary approach which incorporates clinical, radiological and morphological data.

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